While autologous bone marrow transplantation (ABMT) for advanced neuroblastoma may offer advantages over conventional chemotherapy, the major cause of treatment failure in neuroblastoma remains relapse or disease progression. The mechanism of relapse following ABMT is unclear, but may be due at least in part, to residual malignant cells in the transplanted autologous marrow. It can be shown, in fact, using very sensitive assays, that 40-50% of patient marrows are positive for tumor involvement after induction therapy for marrow harvest.

These observations have lead clinicians to incorporate ex vivo bone marrow purging, in which an attempt is made to remove residual neuroblastoma cells from the marrow prior to reinfusion, usually using immunomagnetic methods. The investigational device to be used in the proposed study comprises a panel of monoclonal anti-tumor cell antibodies, anti-murine antibody coated paramagnetic microspheres, and a device, including its associated disposables, designed to remove paramagnetic microspheres added to marrow suspensions. Clinical demonstration of device performance (3-4 logs of tumor cell removal) is not possible due to the usually low infiltration of tumor cells into bone marrow intended for transplant, and the limited sensitivity of detection methods. For this reason, an alternative method employing recombinant DNA technology is proposed to provide information about the source of relapse, and the potential clinical benefit of purging in neuroblastoma patients undergoing autologous bone marrow transplantation.

The proposed study will be an uncontrolled trial of patients with a total of 12 patients with Stage D neuroblastoma in first or second marrow remission as assessed by routine histology. Prior to and in concurrence with this proposed protocol, participating institutions will adhere to their own protocols using intensive combination chemotherapy regimens and drug therapies. Patients treated at the participating institution who are eligible for the concurrent institutional protocol will be eligible for the study. The study will be closed to entry after 12 patients have been entered.

The proposal is to transduce neomycin resistance marker genes, contained in two closely related but distinguishable vectors, into two aliquots of marrow obtained for ABMT. One aliquot of marked marrow will be immunomagnetically purged, then mixed with the second marked, unpurged aliquot and reinfused into the patient. (A third nontransduced backup aliquot will be stored for all patients.) If relapse occurs, the neuroblastoma cells will be separated from the marrow by flow cytometry and analyzed for the presence of marker genes using polymerase chain reaction and restriction fragment length polymorphism assays. Clonogenic assays will also be performed and neuroblast colonies will be analyzed for the presence and frequency of each marker gene to determine the clonogenic potential of the marked cells.

This approach appears to be feasible for neuroblastoma since in vitro studies have shown that fresh clonogenic human neuroblastoma cells can be transduced with an efficiency of approximately 1-3%, and that following in vitro transduction of marrow cells, gene marked normal precursors can be detected in vivo.

Based on a marrow relapse rate of 60% in the first year following transplantation, 24 months of monitoring should allow us to obtain information about the safety of the device and the treatment plan, as well as the utility of gene marking for assessment of the efficacy of purging. Long term follow up (up to 14 years following transplantation) will be provided to address concerns related to the safety of gene marking. Data obtained from the proposed study will be used to design additional studies to statistically validate the safety and efficacy of the Neuroblastoma Bone Marrow Purging System for the removal of neuroblastoma cells from autologous marrow prior to transplantation.